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SUPPORTING INFECTIOUS DISEASE RESEARCH

Enterotoxigenic *Escherichia coli* Expression Clone Set, Recombinant in *Escherichia coli*, Plate 3

Catalog No. NR-19792

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Contributor:

Pathogen Functional Genomics Resource Center at the J. Craig Venter Institute

Manufacturer:

BEI Resources

Product Description:

The Enterotoxigenic *Escherichia coli* (ETEC) expression clone set consists of approximately 900 sequence validated clones from *Escherichia coli* (*E. coli*) strains H10407, E24377A and B7A cloned in *E. coli* DH10B-T1 cells. Each open reading frame was constructed, using ligation independent cloning, in vector <u>pMCSG7</u> (a pET21 derivative that contains an N-terminal 6xHis tag; for routine HTP purification). The sequence was validated by full length sequencing of each clone (using 5' and 3' primers; TACTTCCAATCCAATGCG and TTATCCACTTCCAATG, respectively) with greater than 1X coverage and a mutation rate of less than 0.2%. Please refer to Table 1 for more information on the available clones.

Material Provided:

Each inoculated well of the 96-well plate contains approximately 60 μ L of *E. coli* culture (strain DH10B-T1) in Luria Bertani (LB) Broth containing 100 μ g/mL ampicillin supplemented with 15% glycerol.

<u>Note:</u> Production in the 96-well format has increased risk of cross-contamination between adjacent wells. Individual clones should be purified (e.g. single colony isolation and purification using good microbiological practices) and sequence-verified prior to use. BEI Resources cannot confirm or validate any clone not identified on the plate information table.

Packaging/Storage:

NR-19792 was packaged aseptically in a 96-well plate. The product is provided frozen and should be stored at -80°C or colder immediately upon arrival. For long-term storage, the vapor phase of a liquid nitrogen freezer is recommended. Freeze-thaw cycles should be avoided.

Growth Conditions:

Media:

LB Broth or Agar containing 100 µg/mL ampicillin.

Incubation:

Temperature: *E. coli*, strain DH10B-T1 clones should be grown at 37°C.

Atmosphere: Aerobic

Propagation:

- 1. Scrape top of frozen well with a pipette tip and streak onto agar plate.
- 2. Incubate the plates at 37°C for 18 to 24 hours.

Citation:

Acknowledgment for publications should read "The following reagent was obtained through BEI Resources, NIAID, NIH: Enterotoxigenic *Escherichia coli* Expression Clone Set, Recombinant in *Escherichia coli*, Plate 3, NR-19792."

Biosafety Level: 2

Appropriate safety procedures should always be used with this material. Laboratory safety is discussed in the following publication: U.S. Department of Health and Human Services, Public Health Service, Centers for Disease Control and Prevention, and National Institutes of Health. <u>Biosafety in</u> <u>Microbiological and Biomedical Laboratories</u>. 5th ed. Washington, DC: U.S. Government Printing Office, 2009; see <u>www.cdc.gov/biosafety/publications/bmbl5/index.htm</u>.

Disclaimers:

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References:

1. Stols, L., et al. "A New Vector for High-Throughput, Ligation-Independent Cloning Encoding a Tobacco Etch Virus Protease Cleavage Site." Protein Expr. Purif. 25 (2002): 8-15. PubMed: 12071693.



Table 1: Enterotoxigenic E. coli Expression Clone Set, Recombinant in Escherichia coli, Plate 3 (EEXAC)

Clone	Well	Locus ID	Description	ORF Length	Protein Accession Number	Average Depth of Coverage
D000022881	A03	EcB7A_4173_1_738	spermidine/putrescine ABC transporter, permease protein	791	ZP_03030123	4.1985
D000022883	A04	b7a_C12_g47_1_267	conserved hypothetical protein	323	ZP_03026927	3.2632
D000022886	A05	EcB7A_2311_1_756	conserved hypothetical protein	812	ZP_03030117	4.0333
D000022905	B03	EcB7A_1010_1_846	conserved hypothetical protein	899	ZP_03029740	4.0567
D000022908	B04	b7a_C257_g1_1_273	ABC transporter, periplasmic substrate-binding protein	329	ZP_03026948	3.2006
D000022909	B05	EcB7A_4183_1_846	conserved hypothetical protein	899	ZP_03029740	3.4461
D000022911	B06	b7a_C35_g3_1_273	ABC transporter, periplasmic substrate-binding protein	326	ZP_03026948	3.227
D000022913	B07	EcB7A_2518_1_852	membrane-bound lytic murein transglycosylase C	905	ZP_03029730	4.0652
D000022915	B08	b7a_C116_g3_1_276	hypothetical protein EcB7A_0276	329	ZP_03026897	3.2188
D000022918	B09	EcB7A_4155_1_858	gp59	911	ZP_03027307	3.3787
D000022924	B12	b7a_C4_g15_1_276	hypothetical protein EcB7A_0276	329	ZP_03026897	3.2492
D000022927	C02	EcB7A_3401_1_276	hypothetical protein EcB7A_0276	329	ZP_03026897	3.2249
D000022931	C04	b7a_C12_g51_1_279	DNA-binding protein	335	ZP_03026827	3.2388
D000022934	C05	EcB7A_1776_1_951	transcriptional regulator	1004	ZP_03027967	3.8297
D000022936	C06	b7a_C148_g2_1_279	DNA-binding protein	332	ZP_03026827	2.2199
D000022939	C08	b7a_C214_g1_1_282	hypothetical protein	338	NA	3.2189
D000022946	C11	EcB7A_4052_1_975	TDP-D-fucosamine acetyltransferase	1028	ZP_03027928	3.6449
D000022954	D03	EcB7A_4584_67_1071	YciL domain protein	1061	ZP_03028458	4.5146
D000022956	D04	EcB7A_0562_1_291	periplasmic glucan biosynthesis protein	344	ZP_03026825	3.2297
D000022960	D06	EcB7A_1229_1_291	periplasmic glucan biosynthesis protein	344	ZP_03026825	3.2238
D000022962	D07	EcB7A_0742_1_1047	conserved hypothetical protein	1100	ZP_03028468	4.0036
D000022964	D08	EcB7A_2520_1_291	periplasmic glucan biosynthesis protein	344	ZP_03026825	3.2209
D000022968	D10	EcB7A_4284_1_291	periplasmic glucan biosynthesis protein	344	ZP_03026825	2.2209
D000022970	D11	EcB7A_0777_1_1074	conserved hypothetical protein	1127	ZP_03028468	4.5102
D000022971	D12	b7a_C7_g2_1_294	methyl-accepting chemotaxis protein	347	ZP_03026842	3.1988
D000022976	E02	b7a_C91_g5_1_294	methyl-accepting chemotaxis protein	347	ZP_03026842	2.1902
D000022982	E05	EcB7A_1637_88_1200	hydrogenase-2 operon protein	1169	ZP_03029351	3.9196
D000022983	E06	b7a_C44_g12_1_297	nickel-dependent hydrogenase, b- type cytochrome subunit	350	ZP_03026962	3.2343
D000022986	E07	EcB7A_4904_1_1113	putative invasion gene expression up- regulator	1166	ZP_03028490	4.5575

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Product Information Sheet for NR-19792

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Clone	Well	Locus ID	Description	ORF Length	Protein Accession Number	Average Depth of Coverage
D000022989	E09	EcB7A_4733_76_1200	hydrogenase-2 operon protein	1181	ZP_03029351	4.5651
D000022991	E10	b7a_C91_g8_1_297	nickel-dependent hydrogenase, b- type cytochrome subunit	350	ZP_03026962	3.1914
D000022998	F01	EcB7A_4973_1_1188	oxidoreductase, short chain dehydrogenase/reductase family	1241	ZP_03029384	3.3779
D000023007	F06	b7a_C99_g5_1_300	conserved hypothetical protein	356	ZP_03026936	3.2045
D000023010	F07	EcB7A_3342_679_1899	hypothetical protein EcB7A_1899	1277	ZP_03027102	4.2968
D000023014	F09	EcB7A_4179_1_1269	hypothetical protein EcB7A_1269	1322	ZP_03028583	4.1619
D000023018	F11	EcB7A_1790_160_1482	conserved hypothetical protein	1379	ZP_03027283	3.0218
D000023019	F12	EcB7A_0520_1_303	PAP2 family protein	356	ZP_03026903	3.1601
D000023022	G01	EcB7A_3145_1_1398	conserved hypothetical protein	1451	ZP_03028926	3.4431
D000023024	G02	EcB7A_5042_898_1200	hydrogenase-2 operon protein	359	ZP_03029351	3.2006
D000023028	G04	b7a_C133_g2_70_375	hypothetical protein	362	NA	3.2072
D000023032	G06	b7a_C225_g1_1_306	CDP-alcohol phosphatidyltransferase family protein	362	ZP_03026839	3.2017
D000023035	G08	b7a_C1_g30_1_309	phenylacetic acid degradation protein	365	ZP_03026857	3.2192
D000023038	G09	EcB7A_2481_1_1569	efflux transporter, outer membrane factor (OMF) lipoprotein, NodT family	1622	ZP_03027200	4.5037
D000023043	G12	b7a_C6_g2_1_312	beta-ketoadipyl CoA thiolase	368	ZP_03026937	3.2174
D000023046	H01	EcB7A_3462_1156_283 8	catabolite gene activator	1739	ZP_03030187	4.4756
D000023048	H02	b7a_C116_g2_1_315	enoyl-CoA hydratase	371	ZP_03026981	3.1995
D000023050	H03	EcB7A_3144_1_1746	flagellar hook capping protein	1799	ZP_03030259	4.1668
D000023052	H04	b7a_C12_g46_1_315	enoyl-CoA hydratase	371	ZP_03026981	2.2318
D000023055	H06	b7a_C25_g2_1_315	enoyl-CoA hydratase	371	ZP_03026981	3.2075
D000023060	H08	b7a_C53_g8_1_315	enoyl-CoA hydratase	371	ZP_03026981	3.221
D000023064	H10	EcB7A_4236_1_321	phenylacetyl-CoA oxygenase, alpha subunit	377	ZP_03026919	3.1963